

KOLWI2VH-2:

XXXX	----	N	F	D	----	GV	F	A	-----	XXXXXX	----	TP
SVKG	RF	T	S	R	D	N	S	K	SL	Q	ST	WG
SVKG	----	N	F	D	----	GV	Y	C	-----	DDYGGQ	----	TP
SVKG	----	N	F	D	----	GV	-----	-----	-----	DDYGGQ	----	TP
SVKG	----	N	F	D	----	GV	-----	-----	-----	DDYGGQ	----	TP

FIG. 1

REI:

RATWI2VK:

REIWI2VKRS:

REIWI2VK:

-----M-----S--S--V-----	XXXXXXXXXXXXXX	-	Y	--T--A-K----	XXXXXXXXXX
DIQLTQSPASLPASLGDRVTITC	RASQDIGNYLR	W	F	QQKPGKSPRLIY	GATNLAA
-----M-----S--S--V-----	RASQDIGNYLR	-	-	--T--A-K----	GATNLAA
-----M-----S--S--V-----	RASQDIGNYLR	-	-	--T--A-K----	GATNLAA

REI:

RATWI2VK:

REIWI2VKRS:

REIWI2VK:

-----G	--T	-Y	TF--SS-QP--I-T--		XXXXXXXXXX	--Q	---QI	T-T
GVPSRFGS	RSG	SDF	SLTINSLESDMAIYYC		LHHSEYPYT	FG	I GTKLER	K R
-----G	--T	-	TF--SS-QP--I-T--		LHHSEYPYT	--	---QI	-
-----G	--T	-	TF--SS-QP--I-T--		LHHSEYPYT	--	---QI	-

FIG. 2

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FIG. 3A

oligo 21 cagggtccaactgcaggagtcaggggaggtgtagtgcagcctggaa
[PstI]
CAGGTCCAAC TGCAGGAGTCAGGGGAGGTGTAGTGCAGCCCTGGAAGTCTCTGAGACTT
1 -----+-----+-----+-----+-----+-----+-----+
GTCCAGGTTGACGTCCTCAGTCCCCCTCCACATCACGTCGGACCCTCCAGAGACTCTGAA [oligo
K] 60

Q V Q L Q E S G G V Q P G R S L R L -
|
(Q to V mutation needed)

TCCTGTAGCTCATCTGGATTCACTTACAGTAATTACTGGATGACTTGATACGCCAGGCT
61 -----+-----+-----+-----+-----+-----+-----+
(oligo K) AGGACA TCAGTAGACCTAAGTGAAGTCATTATGACCTACTGAACCTATGCGGTC CGA
120

S C S S S G F T F S N Y W M T W I R Q A -

[KpnI]

CCAGGGAAGGGTCTTGAATGGGTGGTCCATTACTAGTACTGGTGGTGSTACCTACCAT
121 -----+-----+-----+-----+-----+-----+-----+
(oligo K) GGTCCTTCCCAGAACTTACCAACGCAGGTAATGATCATGACCACCACCATGGATGGTA
180
ccaacgcaggtaatgatcatgaccaccaccatggatggtata
P G K G L E W V A S I T S T G G T Y H -

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FIG. 3B

oligo
23 atgcagagtcgtgaaggccgattcactatctccagagataattcaa
[AlwNI]
GCAGAGTCTGTGAAGGCCGATTCACTATCTCCAGAGATAATTCAAAAACACCCGTGTC
181 -----+-----+-----+-----+-----+ 240
CGTCTCAGACACTTCCCGGCTAAGTGATAGAGGTCCTATTAAAGTTTTTGTGGGACAAG [oligo
L]
cgtctcagacactt (oligo 22)
A E S V K G R F T I S R D N S K N T L F -
CTGCAATGGACAGTCTGAGGCCCTGAGGACACGGCGGTTTATTACTGTTCAAGAGATGAC
241 -----+-----+-----+-----+-----+ 300
[oligo L]GACGTTTACCTGTCAGACTCCGGA CTCTGTCGCGCAATAATGACAAGTTCTCTACTG
L Q M D S L R P E D T G V Y C S R D D -
[BstEII]
TACGGAGGACAGACCTATGTTATGGATGCCCTGGGTCAGGGAACCTCCGGTCACCGTC
301 -----+-----+-----+-----+-----+ 360
[oligo L]ATGCCCTCGTCTCGTGATACAATACCTACGGACCCAGTCCCTTGAGGCCAGTGGCAG
caatacctacggaccaccagtccttgaggccagtggcag
Y G G Q S T Y V M D A W G Q G T P V T V -
TCCTCC
361 ----- 366
AGGAGG
aggagg (oligo 24)
S S -

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FIG. 4A

oligo 25 (PvuII)
gacattcagctgaccagtcctccatcttccctgtctgcgtctgtgtgggaga
oligo 25 atgaccagtcctccatcttccctgtctgcgtctgtgtgggaga
GACATTCAGATGACCCAGTCTCCATCTTCCCTGTCTGCGTCTGTGGGAGACAGAGTCACT
1 -----+-----+-----+-----+-----+-----+-----+-----+ 60
CTGTAACTCTACTGGGTCAGAGGTAGAAAGGACAGACGACAGACACCTCTGTCTCAGTGA (oligo
M) D I Q M T Q S P S S L S A S V G D R V T -
ATTACTGCCGGCAAGTCAAGACATTGGAAATTATTTAAGATGGTTCCAGCAGACACCG
61 -----+-----+-----+-----+-----+-----+-----+ 120
[oligo M] TAATGAACGGCCGTTTCAGTCTCTGTAACTTTAATAAATTCTACCAAGGTCGTCGTGTGGC
I T C R A S O D I G N Y L R W F Q Q T P -
oligo 27 tggctgcagggtcccatca
[PstI]
GGGAAAGCTCCGAACTTTTGATTTATGGTGCAACCAACTTGGCTGCAGGGGTCCCATCA
121 -----+-----+-----+-----+-----+-----+-----+ 180
[oligo M] CCTTTCGAGGCTTTGAAAACCTAAATACCACTGGTTGAACCGACGTCCTCCAGGGTAGT
actaaataccacgttggttgaaccgacgtccccagg (oligo 26)
G K A P K L L I Y G A T N L A A G V P S -

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FIG. 4B

cggttcagtgggcagtggtctctggg
 CGGTTCA GTGGCAGTGGGCTCTGGGACAGATTTTACTTTTACCATCTCAAGCCCTTCAGCCT
 181 -----+-----+-----+-----+-----+-----+-----+-----+-----+-----+ 240
 [oligo N] GCCAAGTCACCGTCACCCAGACCCCTGTCTAAATGAAATGGTAGAGTTCGGAAGTCGGA
 R F S G S G S G T D F T F T I S S L Q P -
 GAAGATATTGCTACTTATTACTGTCTGCACCACTTCTGAGTATCCATACACGTTTGGAATT
 241 -----+-----+-----+-----+-----+-----+-----+-----+-----+-----+ 300
 [oligo N] CTTCTATAACGATGAATAATGACAGACGCTGTAAGACTCATAGGTATGTGCAACCTTAA
 E D I A T Y Y C L H H S E Y P Y T F G I -
 GGGACCAAGTTGCAGATCAAAACGTG
 301 -----+-----+-----+-----+-----+-----+-----+-----+-----+-----+ 325
 [oligo N] CCCTGGTTCAACGCTAGTTGCAC
 ccctggttcaacgctctagattgcac (oligo 28)
 G T K L Q I K R -

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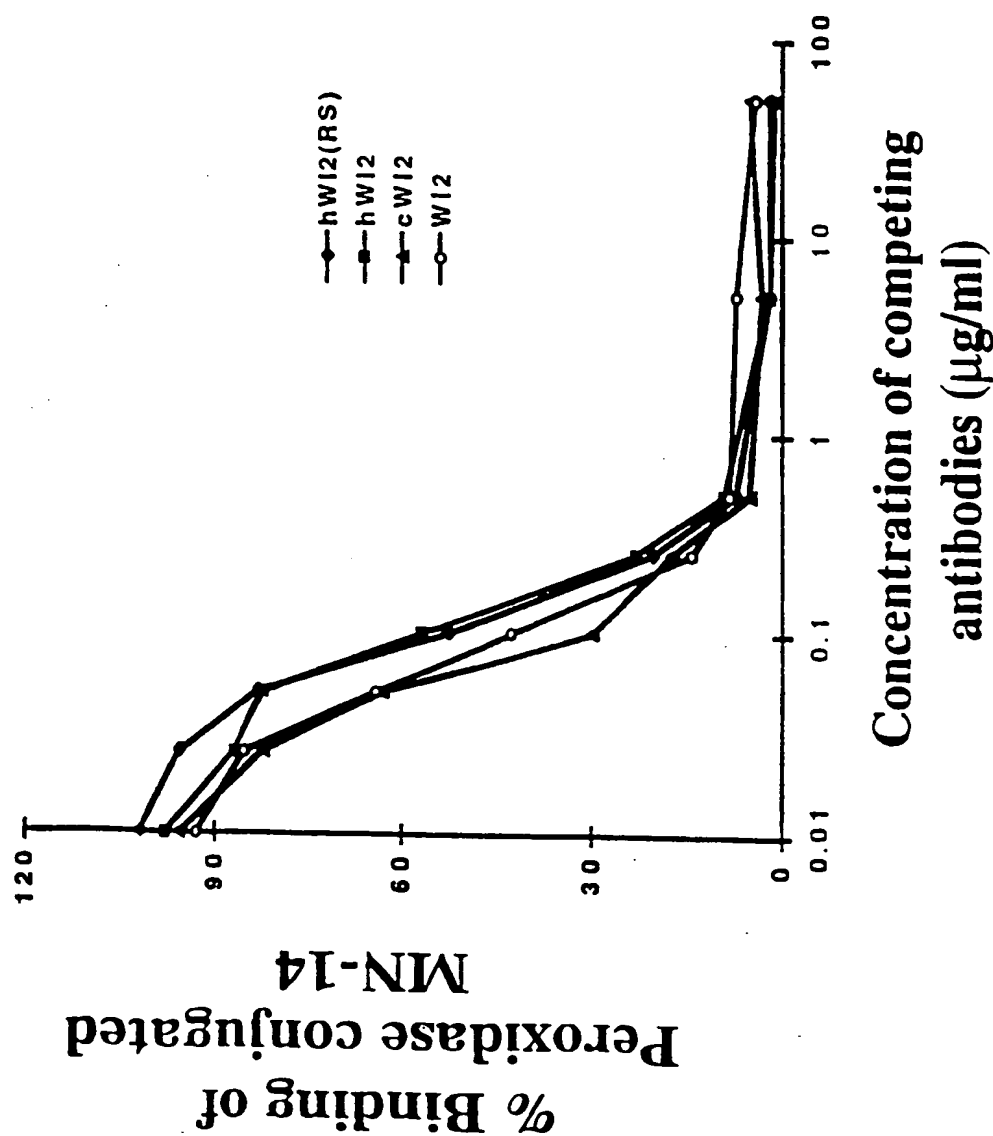


FIG. 5

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GACATTCAGC	TGACCCAGTC	TCCAGCTTCC	CTGCCTGCGT	CTCTGGGAGA	50
CTGTAAGTCG	ACTGGGTCAG	AGGTCTGAAGG	GACGGACGCA	GAGACCCTCT	
D I Q L	T Q S	P A S	L P A S	L G D	
CAGAGTCACT	ATTACTTGCC	GGGCAAGTCA	AGACATTGGA	AATTATTTAA	100
GTCTCAGTGA	TAATGAACGG	CCCGTTTCAGT	TCTGTAACCT	TTAATAAATT	
R V T	I T C R	A S Q	D I G	N Y L R	
CDR1					
GATGGTTCCA	GCAGAAACCG	GGGAAATCTC	CGAGGCTTTT	GATTTATGGT	150
CTACCAAGGT	CGTCTTTGGC	CCCTTTAGAG	GCTCCGAAAA	CTAAATACCA	
W F Q	Q K P	G K S P	R L L	I Y G	
GCAACCAACT	TGGCAGCTGG	GGTCCCATCA	CGGTTTCAGTC	GCAGTAGGTC	200
CGTTGGTTGA	ACCGTCGACC	CCAGGGTAGT	GCCAAGTCAC	CGTCATCCAG	
A T N L	A A G	V P S	R F S G	S R S	
CDR2					
TGGGTCAGAT	TTTTCTCTGA	CCATCAACAG	CCTGGAGTCT	GAAGATATGG	250
ACCCAGTCTA	AAAAGAGACT	GGTAGTTGTC	GGACCTCAGA	CTTCTATACC	
G S D	F S L T	I N S	L E S	E D M A	
CTATTTATTA	CTGTCTGCAC	CATTCTGAGT	ATCCATACAC	GTTTGGAAAT	300
GATAAATAAT	GACAGACGTG	GTAAGACTCA	TAGGTATGTC	CAAACCTTAA	
I Y Y	C L H	H S E Y	P Y T	F G I	
CDR3					
GGGACCAAGC	TGGAACGGAA	ACGG			324
CCCTGGTTTCG	ACCTTGCCTT	TGCC			
G T K L	E R K	R			

FIG.6

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CAGGTCCAAC	TGCAGGAGTC	AGGGGGAGAT	CTAGTGCAGC	CTGGAAGGTC	50
GTCCAGGTTG	ACGTCCTCAG	TCCCCCTCTA	GATCACGTCG	GACCTTCCAG	
Q V Q L	Q E S	G G D	L V Q P	G R S	
TCTGAAACTT	TCCTGTGTAG	CCTCTGGATT	CACATTCAGT	AATTACTGGA	100
AGACTTTGAA	AGGACACATC	GGAGACCTAA	GTGTAAGTCA	TTAATGACCT	
L K L	S C V A	S G F	T F S	<u>N Y W M</u>	
CDR1					
TGACTTGGAT	CCGCCAGGCT	CCAGGGGAGG	GTCTTGAATG	GGTTGCGTCC	150
ACTGAACCTA	GGCGGTCCGA	GGTCCCCTCC	CAGAACTTAC	CCAACGCAGG	
<u>T</u> W I	R Q A	P G E G	L E W	V A <u>S</u>	
ATTACTAGTA	CTGGTGGTGG	GACTTACCAT	GCAGAGTCTG	TGAAGGGCCG	200
TAATGATCAT	GACCACCACC	CTGAATGGTA	CGTCTCAGAC	ACTTCCCGGC	
<u>I T S T</u>	<u>G G G</u>	<u>T Y H</u>	<u>A E S V</u>	<u>K G R</u>	
CDR2					
ATCACTATC	TCCAGAGATA	ATTCAAAAAG	CACCCTGTAC	CTGCAAATGA	250
TAAGTGATAG	AGGTCTCTAT	TAAGTTTTTC	GTGGGACATG	GACGTTTACT	
F T I	S R D N	S K S	T L Y	L Q M N	
ACAGTCTGAG	GCCTGAGGAC	ACGGCCACTT	ATTACTGTTC	AAGAGATAGAC	300
TGTCAGACTC	CGGACTCCTG	TGCCGGTGAA	TAATGACAAG	TTCTCTATCTG	
S L R	P E D	T A T Y	Y C S	R <u>D D</u>	
TACGGAGGAC	AGAGCACCTA	TGTTATGGAT	GCCTGGGGTC	AGGGATCTTC	350
ATGCCTCCTG	TCTCGTGGAT	ACAATACCTA	CGGACCCACG	TCCCTAGAAG	
<u>Y G G O</u>	<u>S T Y</u>	<u>V M D</u>	<u>A W G Q</u>	<u>G S S</u>	
CDR3					
GGTCACCGTC	TCCTCA				6
CCAGTGGCAG	AGGAGT				376
V T V	S S				

FIG.7